

LX.—*On the Formation of the Germinal Layers in Vertebrates.* By BASILIUS LWOFF, of Moscow University*.

EXTENSIVE comparative investigations upon the formation of the germinal layers in Vertebrates have led me to conclude that all the theories at present prevailing on the subject of gastrulation and germinal-layer-formation in this division of the animal kingdom are not in accordance with the facts. All these theories regard the invagination, which is characteristic for all Chordata, as nothing else than gastrulation, although in the majority of cases this invagination bears no relation to the formation of the intestine. The consequence of this is that in the higher Vertebrata we have nothing to help us to distinguish the primary germinal layers. Starting from the conviction that the true endoderm is always formed by invagination, authors have proceeded so far in this direction as to regard the lower layer in the case of the Amniota, from which the intestine is formed and which is homologous with the endoderm of the lower Vertebrates, not as the endoderm, but as something that admits of no homology with what is found in the lower Vertebrates (paraderm &c.); as endoderm, however, are designated the ectoderm cells which are invaginated or grow inwards, but take no part in the formation of the intestine. Or, again, it is assumed that both the cells of the lower layer, from which the intestine is formed, and the invaginated ectoderm cells are to be considered as endoderm; but the lower layer, from which the intestine arises, is designated secondary or cenogenetic endoderm, while to the invaginated cells, which represent the rudiment of the notochord and mesoderm, the term primary or palingenetic endoderm is applied. It therefore follows according to this interpretation that the intestine is formed from the cenogenetic and the notochord from the palingenetic endoderm. In order to realize how untenable this conception is we need only ask which is phylogenetically the older, the intestine or the notochord?

Since we would regard the process of invagination which is characteristic of all Vertebrates as gastrulation in its original simplicity, it is natural that we should desire to discover a gastrula mouth in these supposed gastrulæ also, and to determine in all Vertebrates the homology of the dorsal and ventral lips of the blastopore. But the views of

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different investigators with reference to these questions are so very divergent as to furnish the best proof of the vagueness of the conceptions as to what is to be designated as gastrulation. Just as many differences of opinion prevail on the subject of the gastrula mouth. According to the view of certain embryologists the blastopore in meroblastic ova corresponds to the margin of the epibolic growth which surrounds the yolk. Other embryologists, on the contrary, insist that the margin of the epibolic growth in no way corresponds to the blastopore; they maintain that it is a peculiarity of meroblastic ova, and so forth. It is proposed by this school to designate as blastopore that spot in the germ at which an invagination of cells takes place (in the Selachians the posterior portion of the margin of the germinal disk, in the Amniota the primitive streak and the primitive groove). But this is not all. There are also embryologists who assume that the blastopore is always turned towards the neural side of the animal, where it closes up along a median line, which is termed the gastrula suture ("Gastrularaphe"). In this manner, according to this conception, the nervous system develops in the place of the gastrula suture, since the margins of the blastopore become transformed into the medullary folds!

In this brief communication it is indeed impossible for me to enter into a discussion of all the theories which trace the corresponding developmental processes of Vertebrates to gastrulation, or which, in other words, would discover in gastrulation a universal explanation of the formation of germinal layers, as though no other processes could exist in these stages. Many attempts have been made to apply the gastrulation theory in the case of all Vertebrates, but all these theories are in my opinion forced and unnatural. We have yet to inquire whether it is not possible to interpret these processes somewhat differently without unduly extending the range of the theory of gastrulation.

My investigations have led me to the conclusion that a conception such as regards the process of invagination not as gastrulation, but as a phenomenon which is characteristic of all Chordata, is not merely possible, but is absolutely necessary, if we would compare the earliest developmental processes of different Vertebrates one with another, and at the same time retain the strict homology of the primary germinal layers. I have studied the formation of the germinal layers in the following animals:—*Amphioxus* and *Petromyzon*; *Axolotl*, among the Amphibians; *Pristiurus* and *Torpedo* among the Selachians; *Labrax*, *Julis*, and *Gobius* among Teleostean

fishes; and *Lacerta* among the Reptiles. My investigations upon the development of *Amphioxus* have already been published*. In the following pages it is my intention to describe the results of my studies upon these developmental processes in the above-mentioned Vertebrates, so far as is necessary in order to establish my theory. Although I owe this theory just as much to an attentive study of the literature of the subject as to my own investigations, I shall nevertheless in the present paper leave the literature almost entirely out of the question, in order not to overstep the limits of a provisional communication. The discussion of the literature dealing with the subject, as well as the full description of my investigations, will follow in a detailed paper which is shortly to appear.

Before proceeding to the account of my results I would draw attention to certain points which are of importance in this question. In the first place let us consider whether the school which is at present predominant is correct in regarding the invagination in Vertebrates as gastrulation. In order to answer this question it is necessary to ascertain what is actually to be understood by the term gastrulation, and to discover what portion of the process is typical and what is adventitious and unessential.

Gastrulation is the term which was applied to the process of invagination which leads to the formation of the enteric cavity, whereby a distinct gastrula is constituted. But the process of the formation of the intestine is not always accomplished by means of invagination; it often happens that this process takes place in such a way that the cells which subsequently form the intestine (endoderm cells) are overgrown by the external (ectoderm) cells, and the enteric cavity afterwards arises through divergence of the endoderm cells. This process of enveloping growth is regarded by many investigators as homologous with typical gastrulation, and, as I think, justly, since in both cases the process essentially consists in the fact that the endoderm cells which constitute the intestine come to lie in the interior, and are surrounded by the cells of the ectoderm. Whether this process is accomplished in the form of invagination or of enveloping growth is of subordinate importance. It follows from this that the only process which can be designated as gastrulation is that by which the endodermal elements are invaginated or overgrown, by which consequently above all the formation of the intestine is ushered in. We must hold fast to this con-

* Biol. Centralblatt, Bd. xii. nos. 23 and 24.

ception if we would believe that gastrulation has any definite meaning at all. This is the first point to be considered in this important question.

The second point concerns the discrimination of the primary germinal layers. If we would derive the bilaterally symmetrical Chordata from a Gastrula-like form with radial symmetry we must adhere to the homology of the ectoderm and endoderm of such a Gastrula-like form with the external and internal germinal layers of the Chordata. Since, however, the endoderm of the Gastrula-like form in the first place gives rise to the intestine, our first step in the determination of the germinal layers is to elucidate the question by what elements or what layer the intestine is formed. To this layer the term endoderm must be applied, it matters not whether something besides the intestine is formed from these cells or not.

I will not here enter into the question whether typical gastrulation, *i. e.* invagination, represents a primary or a secondary mode of formation of the endoderm. Nevertheless, in the interpretation of the conditions which are found in Vertebrates I assume that the bilaterally symmetrical Chordata are derived from a Gastrula-like form with radial symmetry, since in the development of the lower Chordata it is impossible not to recognize a gastrula, although a somewhat modified one. At the same time, in homologizing the germinal layers of the Chordata we must not lose sight of our thesis, that the inner layer (the endoderm) of the gastrula forms the intestine, while the ectoderm constitutes the outer covering; otherwise the homologization loses all its meaning. Therefore we shall designate as endoderm cells those from which the intestine arises, no matter whether something else is also formed from these cells or not.

If from this point of view we compare the conditions which exist in *Amphioxus* and the Vertebrates, we arrive at the following results:—The segmentation of the ovum proceeds in such a way that in holoblastic ova, as a result of the segmentation, we get a blastula, one half of which is composed of smaller blastomeres (micromeres) and the other of larger blastomeres (macromeres). The difference between the micro- and macromeres has arisen in consequence of the fact that the former multiply more rapidly than the latter. Since the more rapid multiplication of the micromeres also continues after the formation of the blastula, the micromeres commence to spread out over the macromeres and to grow round them. In cases where we find a single-layered blastula (in *Amphioxus*) this process takes place in such a

way that the macromeres become invaginated; where the blastula consists of several layers (as in *Petromyzon* and Amphibia) the micromeres simply grow round the macromeres. Since these macromeres form the intestine, they may with perfect justice be designated as endoderm cells; the micromeres, on the contrary, which differ noticeably from the macromeres and give rise to the outer covering, are to be termed ectoderm cells. I see no reason to distinguish an ectodermal and an endodermal half after the first divisions of the ovum have taken place, or it may be after the first equatorial furrow (as is the practice of certain investigators). Such a distinction seems to me to be lacking in foundation at this stage, since each supposed endoderm cell of this kind may divide into two or more cells, of which one will subsequently become an ectoderm cell by position, while the other will belong to the endoderm. The distinction of the ectoderm from the endoderm is only possible when the blastula is already formed and the macromeres are being overgrown by the micromeres. In this connexion it makes no difference whether the macromeres are invaginated or are overgrown by the smaller cells. I term them endoderm cells not on account of invagination, but merely because they form the intestine. But the process in consequence of which the endoderm cells come to lie within and become surrounded by the cells of the ectoderm can be homologized with that of gastrulation. This process is to be regarded as preparatory to the formation of the intestine. But besides this process of gastrulation, by which the formation of the intestine is ushered in, there appears on that side which subsequently becomes the dorsal side of the animal another process, which represents the invagination of the ectoderm cells, and which I will term the dorsal invagination. This dorsal invagination is quite independent of the gastrulation, and has nothing to do with the formation of the intestine; it gives rise to the common ectoblastogenous rudiment of the notochord and the mesoderm.

This conception, which was mainly arrived at in the investigation of the development of *Amphioxus*, *Petromyzon*, and *Axolotl*, I was also able to follow out in the interpretation of the developmental processes of the Teleosteans and Selachians, and it is moreover confirmed in the clearest manner by the development of the Amniota.

After these preliminary remarks I will proceed to the account of my results.

My investigations upon the formation of the germinal layers in *Petromyzon* commence with the stage of the blastula. The blastula includes an excentric cavity, lying nearer the upper pole, the roof of which is constituted by smaller blastoderm cells and the floor by several layers of larger cells, richer in yolk, which develop into endoderm. I was also able to observe the multiplication and extension of the blastoderm (ectoderm) cells over those richer in yolk (endoderm cells). Numerous mitoses may be observed in the ectoderm cells. I counted the mitoses in a series of sections through a stage of this kind, and found that the endoderm cells contained five and the superficial ectoderm cells twenty-three mitoses. In this manner smaller ectodermal cells surround the larger endodermal elements, which thus come to lie within and become enveloped by the former. Simultaneously with this on one side, which becomes the dorsal surface of the embryo, a particularly active multiplication of the ectoderm cells may be noticed, and here, at a spot which marks the posterior end of the embryo, the invagination of the ectoderm cells commences. By this means a cavity is formed, which is usually termed the cavity of the gastrula, or archenteron, and the dorsal wall of which is constituted by the invaginated ectoderm cells. But this invagination forms only the dorsal wall of the cavity, and I therefore term it the dorsal invagination. The ventral wall of the cavity is formed by the endoderm cells, which are not invaginated, but were previously situated here. These conditions can be seen with great distinctness in median sections through stages of this kind. In such sections we observe that the ectoderm cells grow inwards from the dorsal margin of the enveloping layer, and form the dorsal wall of the cavity; the ventral wall, on the other hand, is formed by the cells of the endoderm. We also observe the different character of the cells of the dorsal and ventral walls. The former are arranged in epithelial fashion, and form the continuation of the ectoderm cells, which grow inwards from the margin of the enveloping layer. The cells of the ventral wall of the cavity have a roundish or polyhedral form, and are not arranged in epithelial fashion. They acquire the regular epithelium-like character later on, when the intestine arises from them. In the enumeration of the mitoses in a series of sagittal sections it was found that the endoderm cells contained four (chiefly in the anterior portion of the cavity), the ectoderm cells twenty-four (chiefly upon the dorsal side; among these were seven at the margin of the enveloping layer), and the invaginated ectoderm cells four mitoses.

I must lay quite special emphasis upon the fact that the

endoderm cells which subsequently form the intestine are not invaginated; they only experience certain displacements in consequence of the dorsal invagination, whereby the segmentation cavity becomes obliterated. Elements alone are invaginated from which the notochord and the mesoderm arise, and moreover this invagination takes place in such a way that the ectoderm cells grow inwards from the margin of the enveloping layer and form the continuous dorsal plate, the ectoblastogenous rudiment of the notochord and mesoderm. I see no reason for designating the cells of the dorsal wall of the cavity as endodermal, merely because they grow inwards (become invaginated). Similarly I see no reason for terming the cavity at this stage the gastrula or archenteric cavity. At this stage an enteric cavity is not yet present: it is constituted subsequently, when the endoderm cells diverge one from another and form the epithelial wall.

From the central portion of the dorsal plate is differentiated the rudiment of the notochord; the two lateral portions, together with the adjoining cells of the endoderm, form the rudiments of the mesoderm. As a rule no sharp dividing line can be drawn between the ectoblastogenous and endoblastogenous cells of the mesoderm, for the cells which are invaginated grow inwards, while they closely adjoin the cells of the endoderm. After the rudiment of the notochord has separated off from the lateral mesodermal rudiments, the margins of the endoderm grow towards one another, to close the enteric wall; but before this takes place the rudiment of the notochord is temporarily included in the dorsal wall of the intestine. Since the margins of the endoderm continue to grow beneath the notochord, the latter is excluded, the edges of the endoderm unite, and the enteric cavity closed on all sides is now constituted.

I was able to observe the same developmental processes in the case of *Axolotl* also. In this case certain points tell even more in favour of my conception than in *Petromyzon*. While the larger endoderm cells are being overgrown by the smaller ones of the ectoderm the dorsal invagination begins. This invagination is nothing else than the ingrowth of the ectoderm cells, which grow inwards from the margin of the envelope and form the continuous dorsal plate of cells which, as in the case of *Petromyzon*, represents the ectoblastogenous rudiment of the notochord and of the mesoderm. This invagination in no way assists in the formation of the enteric

cavity. The intestine is formed by the divergence and displacement of the endoderm cells, which are not invaginated, but were previously situated here, and may be distinguished all the time from the smaller ectoderm cells by their relative size, by the amount of the yolk-granules which they contain, and to a certain extent by their want of pigment. While the dorsal invagination is taking place, the cavity which subsequently becomes that of the intestine is gradually formed by divergence of the endoderm cells. The formation of the wall of this cavity proceeds in precisely the same manner as in *Amphioxus* and *Petromyzon*, i. e. the endoderm cells separate in such a way that they first give rise to the ventral and lateral boundary of the intestine, while the dorsal portion is still open. Thus it comes to pass that the rudiment of the notochord, which has separated from the lateral mesodermal rudiments, temporarily takes part in the limitation of the enteric cavity. Subsequently the endoderm cells unite beneath the notochord, which in this manner is again excluded. As regards the lateral portions of the dorsal plate-like ingrowth, which represent the ectoblastogenous rudiment of the mesoderm, they are from the first excluded from the delimitation of the enteric cavity owing to the intervening endoderm cells, part of which attach themselves to the ectoblastogenous mesoderm cells in order to furnish their contribution to the formation of the mesoderm.

I must not conceal the fact that these conditions are not exhibited by all Amphibians. According to the statements of certain investigators the Anura are an example of an instance in which the notochord is excluded from the outset from the delimitation of the enteric cavity. Unfortunately I have no experience myself in this direction, since I have not studied the development of the Anura. But if this statement is correct, the Anura in this respect approach the Selachians and Teleosteans, as will be shown further on.

It is sufficiently clear from what has been stated that in the case of the Amphibia also a distinction must be drawn between the dorsal invagination and the formation of the intestine. They are two different processes, which have a certain relation one to another only because the formation in one organism of two neighbouring organs always shows certain points of contact or connexion. As regards the formation of the mesoderm, this arises from ectoderm as well as from endoderm, and moreover not only in the lateral mesodermal rudiments to which reference has already been made, but also in the so-called ventral mesoderm, in which also no sharp dividing line is to be drawn between the ectoblasto-

genous and endoblastogenous mesoderm cells. In this brief communication, however, devoid as it is of figures, I cannot enter more closely into these details, and must postpone an account of them until the publication of the longer paper which is to follow.

In passing on to the description of the corresponding developmental processes in the meroblastic ova of Selachians and Teleostean fishes, I must at the outset join issue with those embryologists who, following the example of Hæckel, would regard the yolk merely as a store-room from which the germ derives nutritive matter, and who would deny to the vitelline elements all participation in the formation of the embryo. Although this view must now be regarded as an anachronism, there are still some investigators even at the present time who assume that the yolk in Teleosteans and Selachians takes no share in segmentation. In contradiction to these embryologists I have to state that, according to the results at which I have arrived, the entire endoderm (the definitive intestine and the endoblastogenous mesoderm) in Teleostean fishes as well as in Selachians owes its origin to the vitelline elements.

I will commence the description of my investigations with the Teleostean fishes, which, owing to the fact that their ova contain less yolk, approach the Amphibians more closely in these processes than do the Selachians, in which the same developmental processes are more modified, probably in consequence of the larger quantity of yolk. In the case of all the Teleosteans studied by me (*Labrax*, *Julis*, and *Gobius*), no sharp boundary can be distinguished between the blastoderm cells and the yolk. The lower blastoderm cells are so intimately connected with the subjacent yolk that no dividing line is to be seen. These cells divide in an equatorial direction, and, moreover, in such a way that the uppermost cell which is thus produced becomes constricted off and joins the blastoderm cells, while the lower one, on the contrary, remains in connexion with the yolk. I have observed no trace of a segmentation cavity in any of the Teleosteans which I have investigated. After the blastoderm has been formed and the blastoderm cells commence to grow round the yolk we may observe upon the surface of the latter a continuous layer of protoplasm with nuclei, around which the outlines of cells are sometimes to be seen. This is the **intermediate layer** of authors, the nuclei of which, which are direct descendants of

nuclei of the lower blastoderm cells, were termed merocytes or periblast nuclei. The presence of so many nuclei without the outlines of cells is probably to be explained as being due to rapid nuclear division without corresponding cell-division. I must lay stress on the fact that these nuclei do not perish, as some investigators would have us believe, but form new cells, which take part in the building-up of the embryo. I shall designate these nuclei simply as **yolk-nuclei**.

After the formation of the blastoderm a more or less distinct boundary may be noticed between the blastoderm and the yolk, or, more strictly speaking, between the blastoderm and the intermediate layer which belongs to the yolk. Yet we may observe all the time the equatorial division of the yolk-nuclei and the formation of new cells, which become constricted off from the yolk and join the blastoderm cells. Now, after the overgrowth of the yolk by the blastoderm cells has commenced it is time to distinguish the primary germinal layers. I consider the entire blastoderm, the cells of which grow over the yolk, as the ectoderm, but the yolk with the yolk-nuclei must be termed the endoderm, since the intestine arises from these vitelline elements.

In passing on to the description of the further developmental processes which usher in the formation of the notochord and the mesoderm it is necessary for me to draw attention to the fact that in none of the Teleosteans which I have examined have I observed an invagination. It is well known that certain investigators maintain that the mesoderm in Teleostean fishes is formed by invagination, while others, on the contrary, assert that it arises by cleavage of the blastoderm cells. Although I myself have not observed any invagination, I think I may assume that no essential difference exists between the two processes as they are described by different investigators; for the invagination in this case also has no connexion with the formation of the intestine, and is nothing else than the forward growth of the reflected edge of the blastoderm. In both cases therefore the same blastodermal or ectodermal elements are engaged, and it is a matter of secondary importance whether the cells are from the outset arranged in such a way that the continuous rudiment of the notochord and mesoderm arises by splitting off from the upper portion of the blastoderm (the rudiment of the nervous system), or whether this rudiment is formed by the forward displacement of the cells starting from the margin of the enveloping layer. Whether by means of the process of splitting-off or by that of so-called invagination, the continuous rudiment of the notochord and mesoderm is equally

formed from the same elements (ectoderm cells) as those from which the nervous system is also developed. In this rudiment the central portion (the rudiment of the notochord) becomes differentiated from the lateral regions, from which the mesoderm arises. But here also it may be seen that the endoderm cells take part in the formation of the mesoderm, since the cells which arise from the yolk-nuclei become constricted off and join the rest of the mesoderm cells. Thus in the present case, as in that of other Vertebrates, we can distinguish a twofold source whence the mesoderm arises (ectoblastogenous and endoblastogenous). While the formation of the notochord and mesoderm is taking place there is formed upon the surface of the yolk from the yolk-nuclei the continuous layer of cells from which the intestine is developed. In this manner in Teleosteans also the intestine does not owe its origin to an invagination, but arises from the derivatives of yolk-nuclei.

While the cells of the blastoderm are growing round the yolk, the yolk-nuclei which are in process of multiplication commence to spread out over the latter, so that soon the entire periphery of the yolk is provided with these nuclei. I must emphatically controvert the view, which is held by many investigators, that these nuclei perish without taking part in the formation of the embryo. It has already been mentioned that the yolk-nuclei give rise to the intestine and participate in the formation of the lateral mesodermal rudiments; but I must add that the nuclei also which are situated in the lower (ventral) portion of the yolk take a share in the formation of the embryo, for in my preparations I notice mitoses in this region, and it may be observed that the cells which are here formed from the yolk-nuclei become constricted off and join the blastoderm cells which have grown round the yolk. I am inclined to assume that these cells are homologous with the ventral mesoderm of the Amphibians. In subsequent stages it may be seen that the yolk-nuclei take part in the formation of the liver, since a portion of the cells which arise from these nuclei is directly transformed into hepatic cells. The importance of the rôle of the yolk-nuclei cannot therefore be open to doubt.

I will now pass on to the Selachians.

Although at the present time Selachians are the favourite subject of embryologists, the latter are nevertheless not agreed as to many important developmental processes in these animals, and, above all, as to the origin of the intestine. While certain investigators regard the intestine as originating through invagination, the rest assert that it is formed from

the yolk-nuclei. On the other hand, while in this manner an important rôle is ascribed to the yolk-nuclei by some investigators, the others deny that they take any share in the formation of the embryo. From my own results I am led to suppose that **not only the intestine, but also the entire endoderm, arises from the yolk-nuclei, while the blastoderm cells represent the ectoderm.** The formation of the intestine from the yolk-nuclei is so distinct in all stages that I cannot understand how this fact can be disputed. But, on the other hand, I must corroborate the assertion that a kind of invagination (blastodermal fold) is to be observed at the posterior margin of the germinal disk. My results consequently reconcile to a certain extent the statements of different investigators, for I confirm both the origin of the intestine from the yolk-nuclei, as also the presence of the invagination. But in the present instance also this invagination or doubling-down of the blastoderm cells has nothing to do with the formation of the intestine. I therefore see in it no process of gastrulation, but, as in the case of other Vertebrates, **the ingrowth of the ectoblastogenous rudiment of the notochord and mesoderm,** which proceeds in a forward direction from the posterior margin of the enveloping layer. The Selachians agree with the Teleostean fishes also in the fact that not only is the inversion of the blastoderm cells to be observed at the posterior margin of the enveloping layer, but also the splitting-off of cells here and there. These two different processes—the formation of the intestine from the yolk-nuclei and the ectoblastogenous invagination—are accomplished simultaneously in such a way that, while the ectoderm cells are invaginated at the posterior margin of the enveloping layer, the continuous sheet of endoderm is formed from the yolk-cells, which give rise to the intestine. The invaginating blastoderm cells in no way constitute the intestinal wall; they merely lie, while they grow inwards, so closely upon the true endoderm cells which have arisen from the yolk-nuclei, that the two different rudiments—the invaginated ectodermal one and the endodermal which has developed on the spot from the yolk-nuclei—are intimately connected one with another. It is the more difficult to draw a dividing line between the two rudiments, since in the lateral rudiments of the mesoderm both kinds of cells are in close connexion.

The rudiment of the notochord is excluded from the limitation of the intestinal cavity by the actual endoderm cells, and forms for a time an appendage of the dorsal wall of the intestine. As regards the mesoderm, we may follow Rückert in distinguishing the axial and peripheral portions of this layer.

In the formation of these parts of the mesoderm, which differ only in position, the cells of both ectoderm and endoderm participate. These two sources of the mesoderm cells are very distinctly visible; but the detailed demonstration must be reserved for my subsequent paper. I will here but briefly allude to one more point. It is evident from what has been stated that I by no means regard the Selachians as primitive forms, from which other animals are to be derived. I see no reason to derive the Amphibians from the Selachians, as is done by certain investigators. This amounts to deriving simple and primitive conditions from such as are complicated and modified. Although the Selachians in many respects exhibit primitive characteristics, their conditions are nevertheless so much modified in consequence of the superabundance of the food-yolk, that they can be regarded as the ancestral type of other animals just as little as the Teleostean fishes.

In the case of *Lacerta* no difficulty is experienced in distinguishing the two primary germinal layers, since we here get a bilaminar germinal disk as the result of segmentation. At the posterior margin of the germinal disk we may observe an active multiplication, and in consequence of this a proliferation of the ectoderm cells. This proliferating spot in the ectoderm, which in the case of Amniota is termed the primitive tubercle ("Primitivknopf"), primitive plate, primitive streak, and I know not what besides, gives rise to the ectoblastogenous rudiment of the notochord and mesoderm, which, starting from this point in a forward direction, grows in between the two primary germinal layers as a continuous plate of cells. In the middle of the primitive streak, but nearer its anterior end, there may be noticed a depression by which the invagination is ushered in. In this manner there arises the margin of the enveloping layer at which the cells of the rudiment of the nervous system and those of the rudiment of the notochord bend round into each other, and where numerous mitoses are to be seen. The direction of these mitoses shows in the clearest manner that the cells of the rudiment of the notochord grow forward from the margin of the overgrowth. This invagination in no way leads to the formation of the intestine, since all the invaginated cells are absorbed in the production of the notochord and mesoderm; but the intestine arises from the cells of the lower primary germinal layer. I have no reason in this case to designate the process

of invagination as gastrulation, for this process here bears no relation to the formation of the intestine. It is true that certain investigators describe a sac-like invagination in the case of Reptiles, and consider its cavity to be that of the archenteron. But, in my opinion, this discovery does not prove what these authors wish to prove, but rather exactly the opposite, since it is evident from their description that, although the cavity produced by invagination exists, nevertheless its walls do not participate in the formation of the intestine, and that the cavity of invagination in no way becomes that of the intestine.

Consequently here also we see in the invagination a process characteristic of the Vertebrata, namely the formation of the ectoblastogenous rudiment of the notochord and mesoderm. In this case also the notochordal rudiment, after it has separated from the lateral rudiments of the mesoderm, enters into a temporary connexion with the endoderm cells, since it becomes intercalated among them; but subsequently the notochord is once more excluded. A correct description of this process has already been furnished by many investigators.

As regards the mesoderm, we can here distinguish the axial and peripheral regions of this layer. The axial mesoderm develops on both sides of the notochord from the ectoblastogenous rudiment already described; but the cells which split off from the endoderm also take part in the formation of these mesodermal rudiments. The peripheral mesoderm is formed from the endoderm cells. To the peripheral mesoderm likewise belongs that at the posterior margin of the primitive streak, which also arises from the cells of the endoderm.

The following important conclusions result from my investigations:—

(1) In the invagination in the case of *Amphioxus* two different processes are to be distinguished:—(a) The invagination of the endoderm cells which form the intestine (the palingenetic process or gastrulation); (b) the dorsal invagination of the ectoderm cells, which constitutes the ectoblastogenous rudiment of the notochord and mesoderm (the ceno-genetic process).

(2) In all Vertebrates these two processes can be distinguished—the origin of the intestine from the endoderm cells, and the formation of the rudiment of the notochord and mesoderm from a proliferation of the cells of the ectoderm.

In no Vertebrate is the intestine formed by invagination. The endoderm cells are overgrown by those of the ectoderm, and the intestine arises through divergence of the former. In the process of overgrowth a modified gastrulation may be seen. But the further this process extends the more it is suppressed. In meroblastic ova, where the yolk is very large (Selachians, Sauropsida), the process of overgrowth is so greatly modified in consequence of adaptation to the new conditions that gastrulation is practically entirely suppressed. The dorsal ectodermal invagination, on the contrary, exists in all Vertebrates as a characteristic process, which has nothing to do with the formation of the intestine, and is to be regarded as a new formation *, that is, as the ingrowth of the ectoblastogenous rudiment of the notochord and mesoderm. This rudiment develops from the cells of the ectoderm and has a common origin with the rudiment of the nervous system.

(3) In no Vertebrate is a true gastrula to be observed; it is therefore impracticable to search for the gastrula mouth in its proper sense, and to endeavour to discover its dorsal and ventral lips, since the primitive conditions are practically entirely obliterated by the new formations. Accordingly I leave this question out of consideration. I consider all attempts to homologize the dorsal and ventral lips of the gastrula mouth in different Vertebrates as forced and unnatural.

(4) There is only one spot in the germ the homology of which can be traced in all Vertebrates, namely that from which the invagination † of the ectoderm cells proceeds—in other words, where the ingrowth of the ectoblastogenous rudiment of the notochord and mesoderm commences.

(5) The so-called neurenteric canal appears to me in a very different light. It is not a tube of communication between the nervous system and the intestinal tract. Just as the margin of the enveloping layer represents a transitional region between the cells of the rudiment of the nervous system and the rudiment of the notochord, so this canal is to be regarded as the morphological expression of this transition. Therefore it may be regarded as a canal of communication between the tube of the nervous system and the notochordal groove or notochordal cavity, and may be termed the *neurochordal*

* It will be evident from this that I virtually dissent from the customary view, according to which the posterior end of the germ, where the invagination is to be observed, exhibits more primitive conditions.

† In the majority of cases it is really not a true invagination, but merely the ingrowth of the cells or the turning-in of the margin of the enveloping layer.

canal. This canal owes its origin to the genetic connexion between the rudiment of the nervous system and that of the notochord.

(6) The fact that the rudiment of the notochord and of the mesoderm arises as a continuous mass of cells from a rudiment which also gives origin to the nervous system may be turned to account for the purposes of phylogeny. It follows from this that, in attempting to derive the bilaterally symmetrical Chordata from Cœlenterate-like forms with radial symmetry, we must remember that the notochord and the mesoderm situated on either side of it in no way arise from the dorsal portion of the intestine, but from the ingrowth of the proliferating ectoderm cells. Since this ectoblastogenous mesoderm is chiefly utilized for the formation of the musculature, we may apply the term *chordo-muscular* to the entire ectoblastogenous rudiment. Since this rudiment has a common origin with that of the nervous system, the Chordata exhibit a direct connexion with the Annelids, in which, according to the investigations of Kleinenberg, the musculature also arises from the common neuro-muscular rudiment. In my opinion the resemblance is still further increased by the presence of the cephalic shield (or ventral shield, as the case may be) in Annelids, which, in its position and its relation to the nervous system and the muscles and in its structure, is very similar to the notochord. It might then be assumed that this continuous neurochordo-muscular rudiment had already made its appearance in the animals which are to be regarded as the common ancestors of the Chordata and the Annelids. The notochord may have arisen in these forms from an ectodermal rudiment as an axial structure which served for the attachment of the musculature on the one side and of the nervous system on the other. The further development and differentiation of different portions of this continuous ectodermal rudiment might have taken different roads. In the Annelids the notochordal rudiment experienced no further development. Perhaps, nevertheless, in the case of certain Annelids a homologue of the notochord may be proved to exist in the epithelial vesicular tissue*, which, in addition to a certain similarity of structure, also exhibits some resemblance to the notochord in the fact that it serves for the attachment both of the nervous system and of the lateral musculature.

The Ascidiæ have followed another path. In these

* I shall later on make a special communication on the subject of this notochord-like structure in Annelids.

animals the notochord is only found in the tail. We might imagine such an animal as an Ascidian larva to have been developed in the following manner:—The proliferation arising at the posterior end by multiplication of the cells, which represents the chordo-muscular rudiment, instead of growing forwards between the intestine and external covering, as in Vertebrates, formed an appendage in which the notochord and the lateral musculature were differentiated from the common rudiment. It must be pointed out that the lateral muscles are present only on either side of the notochord, *i. e.* only in the tail of the larva; in the body of the Ascidian, where there is no notochord, they are entirely absent. This shows with sufficient clearness that the lateral musculature and the notochord here by no means arise from the elements of the endoderm (dorsal portion of the intestine) as is commonly supposed, but, as I think, from a special continuous ectodermal rudiment. In *Amphioxus* and the Vertebrates the proliferation of the ectoderm cells proceeds from the hinder margin in such a way that the chordo-muscular rudiment grows forward between the two primary germinal layers, and thus forms a plate of cells which is situated between the nervous system and the intestine. The ingrowth of this rudiment from the posterior margin of the enveloping layer may be regarded as an ancestral feature, since in the case of the Ascidians also the proliferation takes place at the posterior margin. But this process was subsequently capable of being so far modified that in certain Vertebrates, in addition to growth from the margin of the enveloping layer, splitting-off of the ectoderm cells is also observable. From this point of view the dorsal wall of the gastrula of *Amphioxus*, the dorsal wall of the corresponding stages in *Petromyzon* and the Amphibia, the blastoderm of the Selachians and Teleostean fishes, and the ectodermal portion of the germinal disk with the primitive streak and cephalic process of the Amniota, each represent a continuous rudiment, from which the nervous system, the notochord, and the lateral musculature are developed.

(7) The fact that the notochord, although it arises from an ectoblastogenous rudiment, nevertheless enters into a transient connexion with the cells of the endoderm, may also be turned to account for the purposes of phylogeny. It points to the conclusion that the notochord soon after its origin entered into intimate relations with the intestine, and perhaps also commenced to serve for its attachment. This connexion, however, is to be regarded as being of a secondary nature.

I desire to set up this conception in opposition to the theory of gastrulation which at present prevails. I would mention once more that I reserve for my subsequent paper the full description of my investigations and the detailed statement of proof, as well as the consideration of the literature of the subject. In the present communication I wished to substantiate my view as shortly as possible.

Moscow, Nov. 1892.

LXI.—*Descriptions of some new Longicorn Coleoptera from the Indian Region.* By C. J. GAHAN, M.A., of the British Museum (Nat. Hist.).

[Plate XIX. figs. 4-7.]

THE new species described in this paper form part of some recent accessions to the British-Museum collection. Those from the collections made by Mr. G. Q. Corbett in Burma and by Mr. A. Bell in South India were presented through the intermediation of Dr. Andrewes; while most of the remaining species described are from the collections made by the late Mr. E. T. Atkinson.

Æolesthes basicornis, sp. n.

Aureo-sericeo pubescens; prothorace lateraliter in medio subangulato et leviter tuberculato; dorso transversim undulateque rugoso, versus medium sulcis duabus antice conjunctis longitudinaliter impresso; elytrorum apicibus anguste truncatis, utrisque bidentatis; antennis quam corpore triplo longioribus, articulis 1^o 3^oque transversim rugulosis, hoc apice subabrupte crassato.

Long. 38, lat. 10.5 mm.

Hab. Andaman Islands (*Atkinson Collection*).

The underside of the head is crossed by a deep intergenal groove, which is followed by a second shallower and somewhat ill-defined groove; both grooves are slightly bowed forwards in the middle. The prothorax is somewhat constricted anteriorly; it is obtusely angulate and provided with a very small tubercle at the middle of each side; it is wrinkled above over almost the whole surface, the rugæ having a wavy course in a general transverse direction; two longitudinal impressions, bending towards the middle line to unite anteriorly, cut off a central space which is wrinkled like the rest